

TCA protein precipitation protocol

(originally from Luis Sanchez)

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Stock Solutions: 100% (w/v) Trichloroacetic acid (TCA)
recipe: dissolve 500g TCA (as shipped) into 350 ml dH₂O, store at RT.
(for details, check Maniatias under TCA ppt)

Precipitation Protocol:

1. Add 1 volume of TCA stock to 4 volumes of protein sample.
i.e. in 1.5ml tube with maximum vol., add 250µl TCA to 1.0ml sample.
2. Incubate 10 min at 4°C.
3. Spin tube in microcentrifuge at 14K rpm, 5 min.
4. Remove supernatant, leaving protein pellet intact. Pellet should be formed from whitish, fluffy ppt.
5. Wash pellet with 200µl cold acetone.
6. Spin tube in microfuge at 14K rpm, 5min.
7. Repeat steps 4-6 for a total of 2 acetone washes.
8. Dry pellet by placing tube in 95°C heat block for 5-10 min to drive off acetone.
9. For SDS-PAGE, add 2X or 4X sample buffer (with or without ME) and boil sample for 10 min in 95°C heat block before loading sample onto polyacrylamide gel.